Progressive Shrinkage of the Thalamus Following Middle Cerebral Artery Occlusion in Rats

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Permanent middle cerebral artery occlusion in rats results in infarction in the ipsilateral cortex and caudate nucleus-putamen. In this ischemia model, severe shrinkage of the ipsilateral half of the thalamus was observed several months after surgery. We examined the serial profile of this phenomenon in 40 rats at intervals from 2 weeks to 6 months after the operation. The area of the ipsilateral half of the thalamus as a percentage of the area of the contralateral half was 87% at 2 weeks, 77% at 1 month, 54% at 3 months, and 54% at 6 months. Such severe morphologic change distant from the original ischemic focus has not been reported in models of experimental focal ischemia. Retrograde degeneration is thought to play an important role in this phenomenon. (Stroke 1990;21:1485-1488)

Focal cerebral ischemia causes neuronal injury and morphologic alterations where blood flow reduction is most intense. Once blood flow decreases to below a certain level, neuronal damage usually progresses within a few hours. Thus, the concept of an ischemic blood flow threshold has developed, and morphologic change is considered to be a criterion for determining whether individual neurons will survive or succumb to the ischemic insult.1,2 In some regions of the brain, particularly after brief transient ischemia, ischemic damage develops more slowly; 5–30 minutes of forebrain ischemia causes delayed neuronal death, which is most typically seen in CA1 pyramidal cells in the hippocampus.3,4 Even if this slow damage is considered, the direct injury is completed within a few days following the ischemic insult.

Permanent unilateral middle cerebral artery (MCA) occlusion in rats is now widely used as a model of focal cerebral ischemia.5 This procedure causes infarction in the ipsilateral cerebral cortex and caudate nucleus–putamen complex. Following MCA occlusion in this model, we found that the ipsilateral half of the thalamus progressively shrank during several months. This phenomenon is considered to be a slow type of neuronal damage that follows a long time course. The purpose of this research is to examine the serial profile of shrinkage of the thalamus after unilateral MCA occlusion in rats.

Materials and Methods

We anesthetized 70 male Sprague-Dawley rats, 9–10 weeks old and weighing 320–350 g, with 2% halothane. In 60 rats the proximal part of the left MCA was exposed by the transretro-orbital approach, which was modified from our original method6 for chronic experiments. In 40 rats the left MCA was then electrocauterized using microsurgical techniques and permanently occluded. In 20 sham-operated rats the MCA was only exposed. The remaining 10 rats were not operated on.

The 40 MCA-occluded rats were killed at 2 weeks or 1, 3, or 6 months (n=10 at each time) following MCA occlusion. The 20 sham-operated rats were sacrificed at 2 weeks or 1, 3, or 6 months (n=5 at each time) after surgery. The 10 rats that had not been operated on were also killed as normal controls. Fixation was performed by transcardiac perfusion of 500 ml 3.5% paraformaldehyde in 0.1 M phosphate buffer (pH 7.4) at a pressure of 130 cm H2O. Perfusion-fixed rats were left overnight at 4°C. Their brains were removed the following morning and kept in the same fixative for several days.

The brains were embedded in celloidin, and 25-μm-thick coronal sections located 3.8 mm posterior to the bregma were obtained. Sections were stained with hematoxylin and eosin and photographed.
through a low-power microscope. By means of a computer digitizer (Graphtec KD4030) the areas of the right and left halves of the thalamus (Figure 1) were measured. From this, the left:right ratio was calculated. Values are expressed as mean ± standard deviation. The statistical significance of differences between the MCA-occluded and sham-operated groups was evaluated using Student’s t test.

Results

In the MCA-occluded rats, infarction was seen in the ipsilateral cortex and caudate nucleus-putamen complex. This pattern of ischemic damage is similar to that described originally by Tamura et al.5 In the left half of the thalamus, neuronal necrosis was seen 2 weeks after MCA occlusion. Later, progressive shrinkage of the entire left half of the thalamus was observed (Figure 2). Massive gliosis was seen in the shrinking thalamus. No infarction was seen in the sham-operated and normal control rats.

The left:right ratio was 1.00±0.044 in the normal control group. In the sham-operated groups, the ratio was 0.98±0.061 at 2 weeks, 1.00±0.038 at 1 month, 0.99±0.047 at 3 months, and 0.99±0.041 at 6 months. In the MCA-occluded groups, the ratio was 0.87±0.062 at 2 weeks, 0.77±0.071 at 1 month, 0.54±0.107 at 3 months, and 0.54±0.136 at 6 months (Figure 3). A significant difference was recognized between the sham-operated and MCA-occluded groups (p<0.001).

Discussion

Our results demonstrate that the ipsilateral half of the thalamus progressively shrank during several months after unilateral permanent MCA occlusion in rats. The left half of the thalamus showed a remarkable shrinkage, to nearly half the size of the right half, after 6 months. Because the shrinkage was triggered by left MCA occlusion, this phenomenon might be secondary ischemic neuronal damage that takes a long time to develop. The most important difference between the thalamic atrophy shown here and early ischemic neuronal damage is that tissue damage is never seen in the thalamus during the...
acute stage of ischemia. There exist several possible mechanisms for shrinkage of the thalamus.

First, we must consider local blood flow in the thalamus. Unilateral permanent MCA occlusion in rats produces a consistent focal ischemic lesion in the caudate nucleus—putamen and overlying cortex. Local blood flow in the thalamus decreases slightly in MCA-occluded rats. As measured by Tamura et al using quantitative autoradiography, local blood flow in the ventral thalamus 3 minutes after unilateral MCA occlusion was 1.00±0.14 ml/g/min on the ischemic side and 1.35±0.15 ml/g/min on the contralateral intact side. Blood flow on the ischemic side is greater than the threshold for ischemic neuronal damage, and the thalamus is free from definite neuronal injury during the acute phase of ischemia. Although local blood flow in the thalamus has been studied only during the acute phase, it is unlikely that atrophy of the thalamus was caused by reduced local blood flow itself.

Neuronal cell damage can develop not only via direct insult to the cell body, such as ischemia, but also due to degeneration (retrograde or anterograde) of the axon. Neuroanatomic experiments have used this phenomenon to investigate the nerve fiber connections in the nervous system. Unilateral cortical ablation causes shrinkage of the ipsilateral half of the thalamus. Following aspiration of the limbic, striate, and somatosensory cortex in rabbits, all neurons in the thalamus gradually disappear from the fourth through the 34th postoperative week. These experimental results coincide quite well with our observations. Infarction in the cerebral cortex most likely has the same effect as cortical ablation on the thalamic neurons because both insults destroy neuronal connections between the thalamus and cortex. Retrograde (and to some extent anterograde) degeneration is thought to play an important role in the degeneration of the ipsilateral half of the thalamus following cortical damage regardless of the initial insult (surgical destruction or ischemic injury). Cortical transplants have been reported to reduce thalamic atrophy following cortical ablation in neonatal animals. Axons of cortical neurons degenerate after ablation, but thalamic axons survive and establish connections with transplants placed within cortical lesions. Without this connection between thalamic axons and cortical grafts, thalamic axons degenerate and thalamic neurons die, resulting in thalamic shrinkage. Therefore, thalamic atrophy is mainly due to retrograde degeneration. The fact that atrophy is ameliorated by transplantation suggests that some trophic support is needed for the survival of thalamic neurons.

Focal brain lesions induce changes in blood flow and glucose metabolism in areas remote from the original lesion, an effect called diaschisis. This phenomenon is believed to develop secondary to interruption of the neuronal connections and to the ensuing metabolic suppression. Because rich nerve fiber connections exist between the thalamus and the cerebral cortex, diaschisis can occur in the thalamus following infarction of the cerebral cortex. However, the concept of diaschisis does not imply the irreversible morphologic change that we observed. The influence of focal cerebral ischemia does not seem to be restricted to the primary ischemic focus. Via the neuronal network, the effect of focal ischemia can spread to remote regions. Our results demonstrate a typical and severe form of the long-term distant effect of focal cerebral ischemia.

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References


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